Serial No. 10/601,140

Filed: June 20, 2003 Preliminary Amendment

Page 19 of 40

This listing of claims will replace all prior versions, and listings, of claims in the application:

Listing of Claims:

 (amended) A method for detecting and/or isolating a <u>target</u> nucleic acid molecule having a homopolymeric sequence comprising:

treating a sample containing nucleic acid <u>molecules</u> [compounds] with an LNA oligonucleotide to thereby detect and/or isolate a nucleic acid molecule having said homopolymeric sequence

2. (amended) A method for detecting and/[orisolating] or isolating a target nucleic acid molecule having a repetitive element comprising:

treating a sample containing nucleic acid <u>molecules</u> [compounds] with an LNA oligonucleotide to thereby detect and/or isolate a nucleic acid molecule having the repetitive element.

 (original) A method for detecting and/or isolating a <u>target</u> nucleic acid molecule having a conserved nucleotide sequence comprising:

treating a sample containing nucleic acid molecules [compounds] with an LNA oligonucleotide to thereby detect and/or isolate a nucleic acid molecule having the conserved nucleotide sequence

- 4. (original) The method of any one of claims 1 to 3 wherein a sample comprising the nucleic acid molecules is treated with a lysing buffer comprising a chaotropic agent to lyse cellular material in the sample.
- (amended) The method of any one of claims 1 to 3[4] wherein the LNA oligonucleotide [capture probe] is covalently attached to a solid support.

Serial No. 10/601,140 Filed: June 20, 2003 Preliminary Amendment

Page 20 of 40

6. (amended) The method of any one of claims 1 through 3[5] wherein the LNA oligonucleotide [capture probe] is synthesized with an anthraquinone moiety and a linker at the 5'-end or the 3'-end of said [probe] oligonucleotide.

- 7. (amended) The method of claim 6 wherein said linker is selected from the group consisting of [comprising] one or more of a hexaethylene glycol monomer, dimer, trimer, tetramer, pentamer, hexamer, or higher hexaethylene glycol polymer; a poly-T sequence of 10-50 nucleotides in length; [or] a poly-C sequence of 10-50 nucleotides in length or longer[;]; and [or] a non-base sequence of 10-50 nucleotide units in length or longer.
- 8. (amended) The method of claim 5 wherein said solid support is a polymer support selected from the group consisting of a microtiter plate, polystyrene beads, latex beads, a polymer microscope slide or a polymer-coated microscope slide [or] and a microfluidic slide.
- 9. (amended) The method of claim 1 wherein the LNA oligonucleotide [capture probe\ is complementary to a homopolymeric nucleotide comprising at least about one nucleobase that is different than the bases comprising the homopolymeric nucleic acid sequence.
- 10. (amended) The method of any one of claims 1 through 3[4] wherein the LNA oligonucleotide comprises at least about five repeating consecutive nucleotides.
- 11. (amended) The method of any one of claims 1 through 3[4] wherein the LNA oligonucleotide comprises at least about ten repeating consecutive nucleotides.
- 12. (amended) The method of any one of claims 1 through 3[4] wherein the LNA oligonucleotide comprises at least about twenty to twenty-five repeating consecutive nucleotides.

Attorney Docket No. 57764 (71994) Serial No. 10/601,140 Filed: June 20, 2003

Preliminary Amendment Page 21 of 40

- 13. (amended) The method of any one of claims 1 through 3[4] wherein the LNA oligonucleotide comprises at least about thirty repeating consecutive nucleotides.
- 14. (amended) The method of any one of claims 1 through 3[4] wherein the LNA oligonucleotide comprises at least about forty repeating consecutive nucleotides.
- 15 (amended) The method of any one of claims 1 through 3[4] wherein the LNA oligonucleotide comprises at least about fifty repeating consecutive nucleotides.
- 16. (amended) The method of any one of claims 1 through 3[15] wherein the LNA oligonucleotide molecule is complementary to a nucleotide sequence consisting substantially of a poly(A) nucleotide sequence.
- 17. (amended) The method of claim 15, wherein said LNA oligonucleotide [probe\ is synthesized with an anthraquinone moiety and a linker and at [the] its 5'-end [of said probe], where said linker is selected from the group comprising one or more of a hexaethylene glycol monomer, dimer, trimer, tetramer, pentamer, hexamer, or higher hexaethylene glycol polymer; a poly-T sequence of 10-50 nucleotides in length or a poly-C sequence of 10-50 nucleotides in length or longer; or a non-base sequence of 10-50 nucleotide units in length or longer; and a covalent coupling onto a solid polymer support of said LNA oligonucleotide [probe] is carried out via excitation of the anthraquinone moiety using UV light.
- 18. (amended) The method of any one of claims 1 through 3[15] wherein the LNA oligonucleotide molecule is complementary to a nucleotide sequence consisting substantially of a poly(T) nucleotide sequence.
- 19. (amended) The method of any one of claims 1 through 3[15] wherein the LNA oligonucleotide molecule is complementary to a nucleotide sequence consisting substantially of a poly(G) nucleotide sequence.

Attorney Docket No. 57764 (71994) Serial No. 10/601,140 Filed: June 20, 2003 Preliminary Amendment Page 22 of 40

- 20. (amended) The method of any one of claims 1 through 3[15] wherein the LNA oligonucleotide molecule is complementary to a nucleotide sequence consisting substantially of a poly(U) nucleotide sequence.
- 21. (amended) The method of any one of claims 1 through 3[9] wherein the LNA oligonucleotide molecule is complementary to a nucleotide sequence consisting substantially of a poly(C) nucleotide sequence.
- 22. (amended) The method of claim 18, wherein the LNA oligonucleotide [molecule] is selected from the following table:

Comp. No.	Oligo Name:	Sequence 5'-:	
2	LNA_2.T	5'-biotin-TtTtTtTtTtTtTtTtTtTt (SEQ ID NO: 2)	
3	LNA_3.T	5'-biotin-TttTttTttTttTttTttTtt (SEQ ID NO: 3)	
4	LNA_T ₁₀	5'-biotin-TTTTTTTTT (SEQ ID NO: 4)	
5	LNA_T ₁₅	5'-biotin-TTTTTTTTTTTTT (SEQ ID NO: 5)	
6	LNA_4.T	5'-biotin-ttTtttTtttTtttTtt(SEQ ID NO: 6)	
7	LNA_5.T	5'-biotin-tttTttttTttttTttttTt (SEQ ID NO: 7)	
8	LNA_T ₂₀	5'-biotin-TTTTTTTTTTTTTTTTTT (SEQ ID NO: 8)	
9	LNA_TT	5'-biotin-ttTTtttTTtttTTtt (SEQ ID NO: 9)	
10	LNA_TTT	5'-biotin-ttTTTttttTTTttttTTTt (SEQ ID NO: 10)	

Attorney Docket No. 57764 (71994) Serial No. 10/601,140 Filed: June 20, 2003

Page 23 of 40

Preliminary Amendment

23. (amended) The method of claim 18, wherein the LNA oligonucleotide molecule is selected from the following table:

Comp. No.	Oligo Name:	Sequence 5'-:
11	AQ-HEG₃-2.T	AQ-HEG ₃ -TtTtTtTtTtTtTtTtTtTt (SEQ ID NO: 2)
12	AQ-t15-2.T	AQ-t15-TtTtTtTtTtTtTtTtTtTt (SEQ ID NO: 12)
13	AQ-c15-2.T	AQ-c15-TtTtTtTtTtTtTtTtTtTtTt (SEQ ID NO: 14)
14	AQ-t10-NB5-2.T	AQ-t10-NB5-TtTtTtTtTtTtTtTtTtTt (SEQ ID NOS 15 & 2, respectively)

wherein AQ refers to anthraquinone, HEG refers to hexa-ethylene glycol, t15 (SEQ ID NO: 16) refers to 15-mer deoxy-thymine, c15 (SEQ ID NO: 17) refers to 15-mer deoxy-cytosine, t10-NB5 (SEQ ID NO: 15) refers to 10-mer deoxy-thymine 5-mer non-base, and t refers to DNA thymine and T[:] refers to LNA thymine.

- 24. (amended) The method of claim 23, wherein the LNA oligonucleotide [molecule] is selected from the group of oligonucleotides corresponding to Compounds 2 to 10 herein having an anthraquinone in the 5' position instead of biotin.
- 25 (amended) The method of claim 18, wherein the LNA oligonucleotide [molecule] is selected from the group consisting of a oligonucleotides corresponding to Compounds 2 to 18 herein having an anthraquinone in the 5' position and a linker which is selected from the group [comprising] consisting of one or more of a hexaethylene glycol monomer, dimer, trimer, tetramer, pentamer, hexamer, or higher hexaethylene glycol polymer; a poly-T sequence of 10-50 nucleotides in length [or] and a poly-C sequence of 10-50 nucleotides in length or longer.
- 26. (amended) The method of claim 18, wherein the LNA oligonucleotide molecule is selected from the group consisting of [a] oligonucleotides corresponding to Compounds 2 to 10 herein without the biotin substitution in the 5' position.

Serial No. 10/601,140 Filed: June 20, 2003

Preliminary Amendment

Page 24 of 40

- 27. (amended) The method of claim 2 wherein the LNA oligonucleotide [capture probe] is complementary to a repetitive nucleotide sequence comprising at least about one nucleobase that is different than the bases comprising the repetitive sequence.
- 28. (amended) The method of claim 3 wherein the LNA oligonucleotide [capture probe] is complementary to a conserved nucleotide sequence comprising at least about one nucleobase that is different than the bases comprising the conserved nucleic acid sequence.
- 29. (amended) The method of anyone of claims 1 through 3[14], wherein the LNA oligonucleotide comprises at least one nucleotide having a nucleobase that is different from the nucleobases of the remaining oligonucleotide sequence.
- 30. (amended) The method of any one of claims 1 through <u>3</u>[20] wherein the −1 residue of the LNA oligonucleotide molecule 3' and/or 5' end is an LNA residue.
- 31. (amended) The method of any one of claims 1 through 3[23] wherein the LNA oligonucleotide comprises at least about one or more alpha-L LNA monomers.
- 32. (amended) The method of any one of claims 1 through 3[23] wherein the LNA oligonucleotide comprises at least about one or more xylo-LNA monomers.
- 33. (amended) The method of any one of claims 1 through 3[31] wherein the LNA oligonucleotide [molecule] comprises at least about 20 to 50 percent LNA residues based on total residues of the LNA oligonucleotide.
- 34. (amended) The method of any one of claims 1 through 3[33] wherein the LNA oligonucleotide comprises at least about two or more consecutive LNA molecules.

Serial No. 10/601,140

Filed: June 20, 2003 Preliminary Amendment

Page 25 of 40

35. (amended) The method of any one of claims 1 through 3[34] wherein the LNA oligonucleotide comprises modified and non-modified nucleotide molecules.

36. (amended) The method of any one of claims 1 through 3[35] wherein the LNA oligonucleotide [capture probe] comprises a compound of the formula:

$$5'-Y^{q}-(X^{p}-Y^{n})_{m}-X^{p}-Z-3'$$

wherein X is an LNA monomer, Y is a DNA monomer; Z represents an optional DNA monomer; p is an integer from about 1 to about 15; n is an integer from about 1 to about 15 or n represents 0; q is an integer from about 1 to about 10 or q = 0; and m is an integer from about 5 to about 20.

- 37. (amended) The method of any one of claims 1 through 3[36] wherein the association constant (K_a) of the LNA oligonucleotide is higher than the association constant of [the] complementary strands of a double stranded molecule.
- 38. (amended) The method of any one of claims 1 through $\underline{3}[37]$ wherein the association constant of the LNA oligonucleotide is higher than the disassociation constant (K_d) of the complementary strand of the target sequence in a double stranded molecule.
- 39. (amended) The method of any one of claims 1 through 3[38] wherein the LNA oligonucleotide [capture probe] is complementary to the sequence it is designed to detect and/or isolate.
- 40. (amended) The method of claim 39 wherein the LNA oligonucleotide has at least one base pair difference to [the] a complementary sequence it is designed to detect and/or isolate.

Serial No. 10/601,140

Filed: June 20, 2003 Preliminary Amendment

Page 26 of 40

41. (amended) The method according to claim 40 wherein the LNA oligonucleotide can detect at least about one base pair difference between [the] a complementary polyrepetitive base sequence and the LNA/DNA oligonucleotide.

- 42. (amended) The method of any one of claims 1 through 3[41] wherein the LNA oligonucleotide comprises a fluorophore moiety and a quencher moiety, positioned in such a way that [the] a hybridized state of the oligonucleotide can be distinguished from [the] an unbound state of the oligonucleotide by an increase in the fluorescent signal from the nucleotide.
- 43. (amended) The method of any one of claims 1 through 3[2], wherein the T_m of the LNA oligonucleotide is between about 50°C to about 70°C when the LNA oligonucleotide hybridizes to its complementary sequence.
- 44. (amended) The method of [anyone of] claim[s] 4 [through 43], wherein the chaotropic agent is guanidinium thiocyanate.
- 45. (amended) The method of claim 44 wherein the [concentration of] the guanidinium thiocyanate is at least about 2M.
- 46. (original) The method of claim 44 wherein the concentration of the guanidinium thiocyanate is at least about 3M.
- 47. (original) The method of claim 44 wherein the concentration of the guanidinium thiocyanate is at least about 4M.
- 48. (amended) The method of claim 44 wherein the LNA oligonucleotide hybridizes to the <u>target nucleic acid molecule</u> [repetitive element] at a temperature in the range of 20 65 °C.

Serial No. 10/601,140 Filed: June 20, 2003 Preliminary Amendment

Page 27 of 40

- 49. (amended) The method of claim 48 wherein the LNA oligonucleotide hybridizes to the <u>target nucleic acid molecule</u> [repetitive element] at about 20°C.
- 50. (amended) The method of claim 48 wherein the LNA oligonucleotide hybridizes to the <u>target nucleic acid molecule</u> [repetitive element] at about 37°C.
- 51. (amended) The method of claim 48 wherein the LNA oligonucleotide hybridizes to the <u>target nucleic acid molecule</u> [repetitive element] at about 55°C.
- 52. (amended) The method of claim 48 wherein the LNA oligonucleotide hybridizes to the <u>target nucleic acid molecule</u> [repetitive element] at about 60°C.
- 53. (amended) The method of any one of claims 1 through 3[52] wherein the LNA oligonucleotide [capture probe] is adapted for use as a TaqMan probe or Molecular Beacon.
- 54. (amended) The method of any one of claims 1 through [5]3, wherein the LNA oligonucleotide [capture probe] hybridizes to complementary sequences of eukaryotic RNA.
- oligonucleotide is complementary to [the] poly(A) tails in eukaryotic mRNA and where the said LNA oligonucleotide is synthesized with an anthraquinone moiety and a linker at the 5'-end of said oligonucleotide, wherein said linker is selected from the group consisting of [comprising] one or more of a hexaethylene glycol monomer, dimer, trimer, tetramer, pentamer, hexamer, or higher hexaethylene glycol polymer; a poly-T sequence of 10-50 nucleotides in length; [or] a poly-C sequence of 10-50 nucleotide units in length or longer; [or] and a non-base sequence of 10-50 nucleotide units in length or longer; and [a covalent coupling onto a solid polymer support of] said LNA oligonucleotide [probe] is covalently coupled to a solid polymer support [is carried out] via excitation of the anthraquinone moiety using UV light.

Attorney Docket No. 57764 (71994) Serial No. 10/601,140 Filed: June 20, 2003 Preliminary Amendment

Page 28 of 40

- 56. (amended) The method of claim[s 1 through] 55, wherein the eukaryotic mRNA is isolated using the covalently [attached] <u>coupled</u> LNA oligonucleotide, and detected with nucleic acid probes[, such as DNA, RNA or LNA detection probes], using
 - (i) chemiluminiscence [using enzyme-conjugated nucleic acid probes],
 - (ii) bioluminescence [using firefly or bacterial luciferase or green fluorescent protein as reporter molecule],
 - (iii) ligands incorporated into the nucleic acid probes[, such as digoxigenin (DIG) or fluorescein isothiocyanate (FITC) combined with enzyme-conjugated antiligand antibodies], or
 - (iv) biotin-labeled nucleic acid probes [combined with enzyme-conjugated streptavidin or avidin].
- 57. (amended) The method of claim 56, wherein the eukaryotic mRNA is detected using a[n LNA detection oligonucleotide] <u>nucleic acid probe comprising LNA</u> combined with a tyramide signal amplification system.
- 58. (amended) The method of claim 56, wherein the eukaryotic mRNA is detected using a[n LNA detection oligonucleotide] <u>nucleic acid probe comprising LNA</u>, containing a complementary overhang to a free arm in a dendrimer or a branched oligonucleotide conjugated with several digoxigenin, fluorescein isothiocyanate or biotin molecules or fluorochrome molecules, combined with alkaline phosphatase-conjugated or horse radish peroxidase-conjugated anti-digoxigenin, anti- fluorescein isothiocyanate antibodies or streptavidin or detection of fluorescence from the excited fluorochromes.
- 59. (amended) The method of [any one of claims 1-58] <u>claim 55</u>, further comprising contacting the sample with a polymerase and at least one nucleotide.

Serial No. 10/601,140 Filed: June 20, 2003 Preliminary Amendment

Page 29 of 40

60. (amended) The method of claim 59, further comprising performing said contacting under conditions suitable for generating a plurality of copies of said [nucleic acid molecule] eukaryotic mRNA.

- 61. (amended) The method of claim <u>60</u>[59], wherein said conditions comprise exposing the sample to a constant temperature.
- 62. (amended) The method of claim <u>60</u>[59], wherein said conditions comprise cycling the temperature of the sample.
- 63. (amended) The method of claim <u>60</u>[59], wherein the polymerase comprises a thermally stable polymerase.
- 64. (amended) The method of claim 59 or 63, wherein the polymerase comprises a reverse transcriptase.
- 65. (original) The method of claim 59, wherein the LNA oligonucleotide comprises a label.
- 66. (original) The method of claim 59, wherein the nucleic acid molecule or LNA oligonucleotide is bound to a solid support.
- 67. (original) The method according to claim 59 or 65, wherein the at least one nucleotide comprises a label.
- 68. (original) The method of claim 59, wherein the nucleic acid molecule is comprised with a cell and wherein the cell is stably associated with a solid support.
- 69. (amended) The method of claim 60, wherein the LNA oligonucleotide comprises a fluorescent reporter molecule at one end of the LNA oligonucleotide and a quencher

Serial No. 10/601,140

Filed: June 20, 2003 Preliminary Amendment

Page 30 of 40

molecule at another end of the oligonucleotide, wherein the quencher is in sufficient proximity to the reporter to quench the fluorescence of the reporter [label] molecule.

- 70. (amended) The method of claim <u>60</u>[69], wherein generating the plurality of copies is detected by detecting increased fluorescence of the reporter molecule.
- 71. (original) The method of claim 70, wherein the LNA oligonucleotide is cleaved during the step of generating the plurality of copies.
- 72. (amended) The method of [any one of claims 69 through 71] <u>claim 59</u>, wherein the polymerase is rTh polymerase.
- 73. (amended) The method according to claim 59, further comprising adding at least one primer which hybridizes to a sequence in the nucleic acid molecule 5' or 3' of the homopolymeric sequence.
- 74. (amended) The method of any one of claims 1 through 3[4], wherein the LNA oligonucleotide comprises a fluorescent reporter molecule at one end of the oligonucleotide and a quencher molecule at a second end and wherein the reporter molecule is quenched by the quencher molecule when the LNA oligonucleotide is not hybridized to the nucleic acid molecule.
- 75. (original) The method of claim 74, wherein hybridization of the LNA oligonucleotide is detected by detecting increased fluorescence of the reporter molecule.
- 76. (original) The method of claim 74, wherein the LNA oligonucleotide comprises, in addition to a sequence sufficiently complementary to said nucleic acid molecule to specifically hybridize to said nucleic acid molecule, a first and second complementary sequence which specifically hybridize to each other when the oligonucleotide is not hybridized to the nucleic acid molecule, bringing said

Serial No. 10/601,140

Filed: June 20, 2003 Preliminary Amendment

Page 31 of 40

quencher molecule in sufficient proximity to said reporter molecule to quench fluorescence of the reporter molecule.

- 77. (amended) The method of [any one of] claim[s] 59 [through 66], further comprising adding a DNA polymerase, RNaseH and E. coli DNA ligase after conversion of the eukaryotic polyadenylated mRNA to first strand complementary DNA under conditions suitable for generating double stranded complementary DNA
- 78. (original) The method of claim 77 further comprising cloning of said double stranded DNA molecules into a cloning vector thereby generating a library of double stranded complementary DNAs
- 79. (amended) The method of claim 77 where the LNA oligonucleotide complementary to the poly(A) tail sequence in eukaryotic mRNA contains an anchor sequence for a RNA polymerase[, such as T7 RNA polymerase].
- 80. (original) The method of claim 78 further comprising adding an RNA polymerase, such as T7 RNA polymerase, under conditions suitable for generating a plurality of RNA copies of said nucleic acid molecule.
- 81. (amended) A kit for detecting and/or isolating a nucleic acid molecule in a sample comprising:
 - a. an LNA oligonucleotide comprising a nucleotide [acid] sequence sufficiently complementary to a target nucleic acid molecule which comprises a homopolymeric sequence, a repetitive sequence and/or a conserved sequence, to specifically hybridize to the nucleic acid molecule; and
 - b. a label.
- 82. (amended) The kit of claim <u>81</u>[77], wherein the label is coupled to the LNA oligonucleotide, or to a molecule which is capable of hybridizing to the LNA

Serial No. 10/601,140

Filed: June 20, 2003 Preliminary Amendment

Page 32 of 40

molecule, or to a nucleotide which can be incorporated into a primer extension product comprising the LNA oligonucleotide.

- 83. (amended) The kit of claim <u>81</u>[77], wherein the kit further comprises one or more of a polymerase, at least one nucleotide, at least one primer sequence capable of hybridizing to the nucleic acid molecule or to the LNA oligonucleotide, a buffer, Mg²⁺, UNG, a control nucleic acid molecule, a nuclease, a restriction enzyme, a solid support, a capture molecule for binding the nucleic acid molecule to a solid support, a capture molecule for binding the LNA oligonucleotide to a solid support, a tyramide amplification molecule, a dendrimer, and a chaotropic agent.
- 84. (amended) The kit of [any one of claims 77 through 79] claim 81, wherein the LNA molecule comprises the formula:

$$5'-Y^{q}-(X^{p}-Y^{n})_{m}-X^{p}-Z-3'$$

wherein X is an LNA monomer, Y is a DNA monomer; Z represents an optional DNA monomer; p is an integer from about 1 to about 15; n is an integer from about 1 to about 15 or n represents 0; q is an integer from about 1 to about 10 or q = 0; and m is an integer from about 5 to about 20.

- 85. (amended) The kit of [any one of claims 77 through 79] claim 81, wherein the nucleic acid molecule is a eukaryotic RNA.
- 86. (amended) The kit according to claim 81, wherein the LNA oligonucleotide specifically binds to a poly(A) tail sequence in [the] eukaryotic RNA.
- 87. (amended) The kit of [any one of claims 77 through 79] <u>claim 81</u>, wherein the LNA oligonucleotide is an anchor primer.
- 88. (amended) The kit of [any one of claims 77 through 79] <u>claim 81</u>, wherein the LNA is a [Taqman] <u>TaqMan</u> probe or a molecular beacon.

Serial No. 10/601,140

Filed: June 20, 2003 Preliminary Amendment

Page 33 of 40

89. (amended) The kit of [any one of claims 77 through 79] claim 81, wherein the polymerase is a thermally stable DNA polymerase or a thermally stable reverse

transcriptase.

90. (amended) The method of claim 54, wherein the LNA oligonucleotide [capture

probe] hybridizes to complementary sequences of yeast RNA.

91. (amended) The method of claim <u>54[85]</u>, wherein the LNA oligonucleotide [capture

probe] hybridizes to complementary sequences of mRNA, rRNA, and/or tRNA.

87. (amended) A method for amplifying a target nucleic acid molecule the nucleotide

sequence which is complementary to a LNA oligonucleotide capture probe, the

method comprising:

providing a sample containing nucleic acid molecules having repetitive base

sequences; and,

contacting the nucleic acid[s] molecules [released] from the sample with at

least one LNA oligonucleotide capture probe to capture target nucleic acid

molecules; and,

subjecting the captured nucleic acids to polymerase chain reaction, using

primers to amplify the captured nucleic acid molecules.

88. (original) The method of claim 87 wherein multiple primers are used in multiplex

PCR.

89. (amended) A kit for isolating a target nucleic acid having a repetitive or

homopolymeric base sequence, comprising:

an LNA oligonucleotide complementary to the target nucleic acid; and

a substrate for immobilizing the LNA oligonucleotide.

90. (original) The kit of claim 89 wherein the substrate is a microchip array.

-33-

Serial No. 10/601,140

Filed: June 20, 2003 Preliminary Amendment

Page 34 of 40

91. (amended) The kit of claim 89, wherein the LNA oligonucleotide is complementary to a homopolymeric nucleotide <u>sequence</u> comprising at least about one nucleobase that is different than the bases comprising the homopolymeric nucleic acid sequence.

92. (original) The kit of claim 89, wherein the LNA oligonucleotide comprises at least

about five repeating consecutive nucleotides.

93. (original) The kit of claim 89, wherein the LNA oligonucleotide comprises at least

about ten repeating consecutive nucleotides.

94. (original) The kit of claim 89, wherein the LNA oligonucleotide comprises at least

about twenty to twenty-five repeating consecutive nucleotides.

95. (amended) The kit of claim[s] 92 [to 94] wherein the LNA oligonucleotide molecule

is complementary to a nucleotide sequence consisting substantially of a poly(A)

nucleotide sequence.

96. (amended) The kit of claim[s] 92 [to 94], wherein the LNA oligonucleotide molecule

is complementary to a nucleotide sequence consisting substantially of a poly(T)

nucleotide sequence.

97. (amended) The kit of claim[s] 92 [to 94], wherein the LNA oligonucleotide molecule

is complementary to a nucleotide sequence consisting substantially of a poly(G)

nucleotide sequence.

98. (amended) The kit of claim[s] 92 [to 95], wherein the LNA oligonucleotide

[molecule] is complementary to a nucleotide sequence consisting substantially of a

poly(U) nucleotide sequence.

-34-

Serial No. 10/601,140 Filed: June 20, 2003

Preliminary Amendment

Page 35 of 40

99. (amended) The kit of claim[s] 92 [to 95], wherein the LNA oligonucleotide [molecule] is complementary to a nucleotide sequence consisting substantially of a poly(C) nucleotide sequence.

- 100. (original) The kit of claim 89, wherein the LNA oligonucleotide is substantially homologous to the target nucleic acid sequence.
- 101. (original) The kit of claim 89, wherein the LNA oligonucleotide hybridizes to a target nucleic acid sequence in the presence of a chaotropic agent.
- 102. (original) The kit of claim 101, wherein the chaotropic agent is guanidinium thiocyanate.
- 103. (original) The kit of claim 101, wherein the concentration of the guanidinium thiocyanate is at least between about 2M to about 5M.
- 104. (amended) The kit of claim[s] 89 [to 103] wherein the <u>LNA</u> oligonucleotide hybridizes to the repetitive [element] or <u>homopolymeric sequence</u> at a temperature in the range of between about 20 65 °C.
- 105. (original) A method for isolating RNA from infectious disease[s] organisms wherein the genome of the infectious disease organism is comprised of RNA, <u>said genome</u> comprising a consecutively repeating nucleic acid base, the method comprising:

providing a sample containing genomic RNA; and,

treating the sample with a lysing buffer containing a chaotropic agent to lyse cellular material in the sample, dissolve the components and denature the genomic RNA in the sample; and,

contacting [the] genomic RNA released from the sample with at least one [capturing] LNA oligonucleotide <u>capture</u> probe, wherein[,] the capturing probe

Serial No. 10/601,140

Filed: June 20, 2003 Preliminary Amendment

Page 36 of 40

[being] is substantially complementary to [a] the consecutively repeating nucleic acid base in the genomic RNA.

- 106. (original) The method of claim 105, wherein the chaotropic agent is guanidinium thiocyanate.
- 107. (original) The method of claim 106, wherein the concentration of the guanidinium thiocyanate is between about 2M to about 5M.
- 108. (amended) The method of claim 105 wherein the T_m of the <u>at least one LNA</u> oligonucleotide capture probe when bound to its complementary genomic RNA sequence is between about 55°C to about 70°C.
- 109. (original) The method of claim 105, wherein the genomic RNA is protected from degradation by RNAse inhibitors in the presence of the chaotropic agent.
- 110. (amended) The method of claim 105, wherein the genomic RNA is protected from degradation by RNAse inhibitors when hybridized to the <u>at least one</u> LNA oligonucleotide capture probe.
- 111. (original) The method of claim 109, wherein the genomic RNA is isolated from retroviruses.
- 112. (original) The method of claim 111, wherein the retrovirus is HIV.
- 113. (original) The method of claim 109, wherein the isolated genomic RNA is used to genotype RNA viruses.
- 114. (original) The method of claim 109, wherein the isolated genomic RNA is used for diagnosis of an infectious disease organism in a patient suffering from an infectious disease.

Serial No. 10/601,140

Filed: June 20, 2003 Preliminary Amendment

Page 37 of 40

115. (original) A composition comprising an LNA/DNA mixmer oligonucleotide capture probe wherein the LNA/DNA mixmer comprises at least about ten repeating

consecutive nucleotides.

116. (original) The composition according to claim 115, wherein the LNA/DNA

oligonucleotide mixmer comprises at least about twenty-five repeating consecutive

nucleotides.

117. (amended) The composition according to [any of] claim[s] 115 [or 118], wherein the

LNA/DNA oligonucleotide mixmer is complementary to a poly(G) sequence.

118. (amended) The composition according to [any of] claim[s] 115 [or 118], wherein the

LNA/DNA oligonucleotide mixmer is complementary to a poly(U) sequence.

119. (amended) The composition according to [any of] claim[s] 115 [or 118], wherein the

LNA/DNA oligonucleotide molecule is complementary to a poly(C) sequence.

120. (amended) The composition according to [any of] claim[s] 115 [or 118,], wherein the

LNA/DNA oligonucleotide molecule is complementary to a poly(A) sequence.

121. (amended) The composition according to [any of] claim[s] 115 [or 118,], wherein the

LNA/DNA oligonucleotide molecule is complementary to a poly(T) sequence.

122. (amended) The method of any[one] of claims 1 through [5]3, wherein the detection

and/or isolation of a nucleic acid is carried out under high stringency hybridisation

[condisiton] conditions using low salt concentration, optionally after treating the

sample with a lysing buffer comprising a chaotropic agent.

123. (amended) The method of claim 122 wherein said chaotropic agent is GuSCN in a

concentration of at least 4 M.

-37-

Serial No. 10/601,140 Filed: June 20, 2003 Preliminary Amendment

Page 38 of 40

- 124. (amended) The method of claim 122 [or 123] the method further comprises the step of binding the LNA oligonucleotide to nucleic acids from the sample in a [the] binding buffer [contains] containing NaCl or LiCl.
- 125. (amended) The method of claim 124 where [the] NaCl or the LiCl concentration is less than 100 mM.
- 126. (amended) The method of claim 125 where [the] NaCl or the LiCl concentration is less than 50 mM.
- 127. (amended) The method of claim 125 where<u>in</u> [the] NaCl or [the] LiCl concentration is less than 25 mM.
- 128. (amended) The method of claims 122 through 128 wherein [the] detection or hybridisation is carried out at at least 25 °C.
- 129. (amended) The method of claims 122 through 128 where <u>in</u> [the] detection or hybridisation is carried out at at least 37 °C.
- 130. (amended) The method of claims 122 through 128 where <u>in</u> [the] detection or hybridisation is carried out at at least 50 °C.
- 131. (new) The method according to claim 56 comprising detecting chemiluminiscence using enzyme-conjugated nucleic acid probes.
- 132. (new) The method according to claim 56 comprising detecting bioluminescence using firefly or bacterial luciferase or green fluorescent protein as reporter molecule.
- 133. (new) The method according to claim 56 comprising detecting bioluminescence using firefly or bacterial luciferase or green fluorescent protein as reporter molecule.
- 134. (new) The method according to claim 56 comprising detecting digoxigenin (DIG), fluorescein isothiocyanate (FITC), or biotin incorporated into the nucleic acid probes.

Attorney Docket No. 57764 (71994) Serial No. 10/601,140 Filed: June 20, 2003 Preliminary Amendment Page 39 of 40

135. (new) The method of claim 79 wherein the RNA polymerase comprises a T7 RNA polymerase.